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Short communication

Effects of fire history and N and P fertilization on seedling biomass, Specific Leaf Area, and root:shoot ratios in a South African savannah

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ABSTRACT

Fire exerts clear direct effects on savannah vegetation dynamics, but the indirect effects of fire, for example via changes in soil fertility, are less clear. In fire-dominated nutrient-poor savannahs, there is little evidence that fire affects plant-available nutrients, but plant performance on soils with contrasting fire histories has not been investigated. A bioassay experiment was conducted in Kruger National Park, South Africa, to test for effects of long-term fire manipulation on the ability of soils to influence tree growth. *Combretum hereroense*, a common savannah species, was used as a phytometer species in an experiment that combined fire history with N and P fertilization treatments. The results suggested no effect of fire history on a number of seedling traits after seven months of growth. Root mass and shoot:root ratios responded to P addition, with seedlings exhibiting lower allocation to root tissue under elevated P, particularly in the presence of N, suggesting some N and P co-limitation. Overall, it is concluded that there is little evidence that fire degrades or alters soil chemistry in a way that is likely to influence vegetation structure and dynamics in nutrient-poor savannah ecosystems.

1. Introduction

Fire plays a key role in the ecology of many savannah ecosystems, and the direct effects of fire on plant demography have been quite well documented (Bond, 2008; Higgins et al., 2007; Holdo et al., 2009a, 2009b). The indirect effects of fire on plants, i.e., those mediated through soil processes, are less clear. Fire can alter soil structure and chemistry, with potentially important consequences for nutrient and water cycles and plant uptake of key resources (Blair, 1997; Hartnett et al., 2004; Johnson and Matchett, 2001). Fire has been shown to lead to N depletion in soils in temperate grassland ecosystems (Ojima et al., 1994; Risser and Parton, 1982; Turner et al., 1997), for example, but in savannahs the evidence for such an effect has been conflicting (Coetsee et al., 2008; Fynn et al., 2003). Many savannahs occur on nutrient-poor, sandy soils, and in these systems in particular, there is little evidence for fire effects on plant-available nutrients (Aranibar et al., 2003; Coetsee et al., 2010, 2008; Holdo et al., 2012). So far, there has been little evidence to support the view that fire exerts a significant impact on soil nutrient availability on these soils, even after half a century of fire manipulation (Holdo et al., 2012). Assuming that savannah trees are nutrient-limited, this would suggest that fire is unlikely to influence tree growth through its effects on soil properties. Soil analyses, however, do not necessarily identify the factors limiting plant growth (Axmanová et al., 2011), and to the knowledge of this author no studies have been conducted to date that directly examine the relationship between the fire history of savannah soils and tree growth or other key traits. Fire could plausibly affect soil factors that are important for tree performance that have not been measured (or are difficult to quantify), for example by reducing the size of a key nutrient pool or by disruption of symbiotic relationships between plants and mycorrhizae.

To examine the role of fire history on plant performance through such potentially unmeasured factors, a bioassay experiment (Axmanová et al., 2011; Köhler et al., 2001) was conducted in which seedlings of a common savannah species were grown in soils with contrasting fire histories. A previous study revealed no effect of fire (August annual and triennial burns compared with each other and with fire-protected control plots) on soil total or mineralizable N or Mehlich-P in a long-term (>55 years of experimental manipulation at the time of the study) fire experiment in Kruger National Park (Holdo et al., 2012).

2. Material and methods

2.1. Study site

The study was conducted in Kruger National Park (KNP), a tropical/sub-tropical savannah ecosystem with a strong unimodal rainy season (Holdo et al., 2012). Soils for the bioassay experiment were collected

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at one of the sites corresponding to the Experimental Burning Trial, an ongoing long-term fire experimental manipulation established in 1954 (Biggs et al., 2003; Govender et al., 2006). Each site in the experimental burning trial consists of four 'strings' or blocks (Biggs et al., 2003). Two treatments (season and frequency of burn, in an incomplete design) are applied within plots (on average about 7 ha in size) in each block. Soil samples (to 20 cm depth) were collected from burn plots corresponding to the Napi, Nwashitsaka, and Mbiyamiti blocks belonging to the Skukuza site, which sits on nutrient-poor, sandy soils of granitic origin, in an area receiving about 650 mm of annual rainfall. Only three of the four blocks from the Skukuza site were used because one block is substantially geologically different from the remaining three (Holdo et al., 2012).

2.2. Experimental setup and analysis

In July 2010, soil was collected from the control plot (complete fire suppression), and the annual and triennial August burn plots of each block, resulting in nine total soil sources (three burn treatments replicated three times). Soil collections were conducted in open areas (away from tree canopies) to avoid possible confounding sub-canopy effects, which are known to be important in this system (Coetsee et al., 2010; Holdo et al., 2012). Soil samples were collected in large plastic bags from multiple sites within each plot and returned to the Skukuza Nursery, where they were sifted to remove rocks and roots and combined. The soils were then placed in 4-L containers and subjected to a fertilization treatment at the Nursery, the purpose of which was to (1) determine whether N and P are limiting for tree growth and (2) examine fertilization by soil source interactions to determine whether any fire history effects are most likely to be related to N or P limitation. The containers were large enough to prevent seedlings from becoming pot-bound, and all fertilization treatments were applied on a per-area basis after calculation of pot diameter and horizontal surface area. The soil source ("fire" hereafter) treatment was crossed in a fully factorial design with N and P fertilization treatments, with 4–6 replicates per treatment combination. A single level of N or P augmentation was used. Ammonium nitrate and urea were used for the N fertilization treatment, and superphosphate for P. Fertilizer was applied once at the initiation of the experiment in July 2010, at a rate of 10 g Nm^{-2} and 5 g Pm^{-2}. These fertilization rates matched those used by Craine et al. (2008) in a field fertilization experiment at KNP that resulted in a significant growth response of herbaceous vegetation.

Combretum hereroense Schinz (Combretaceae) was chosen as the phytometer species in the bioassay experiment because it is common in and around the Skukuza burn plots, and has been observed occurring across treatments (control, annual and triennial burns, *pers. obs.*). *C. hereroense* is a species with a broad range, usually encountered as a small tree in open woodland and wooded grassland (Coates Palgrave, 1977). Nursery staff collected seeds from the field and germinated them in the Nursery beginning in August 2010. Seeds were germinated in trays on elevated tables under shade cloth and watered to saturation on a daily basis. Upon germination, seedlings were transplanted to the experimental containers in November 2010. The experiment was conducted outdoors, under shade cloth to prevent seedling desiccation. Water was applied to field capacity on a daily basis and the pots were rotated frequently to reduce the likelihood of light gradients or other factors across the area of the experiment. A first analysis of plant survival was conducted in March 2011 (three months after transplantation) to determine whether transplant failure was related to experimental treatments. Each seedling was scored as live or dead and insect herbivore damage was ranked categorically as "none", "moderate", or "severe". At the end of the experiment in July 2011 (seven months after transplantation), all seedlings were harvested. Three fresh, fully-expanded leaves from each plant were collected, photographed and digitized in the lab with Sigmascan (Systat, San Jose, CA) for area determination. The leaves were then oven-dried in the lab at 60 °C for 48 h. Subsequently, dry mass was determined using a precision (10^{-4} g) scale (Mettler Toledo, Columbus, OH). Specific Leaf Area (SLA) was calculated by dividing the combined leaf surface area by the total biomass of the three leaves. SLA is an important synthetic plant trait that correlated a large number of plant life history strategies and functional traits (Hoffmann et al., 2005; Reich et al., 1998). During harvesting, each plant was carefully washed clean of soil by dipping it in a bucket filled with water. The shoot and root were separated and oven-dried in the lab for dry mass determination.

The final response variables analyzed were SLA, shoot mass, root mass, and root:shoot mass ratio. There was minimal herbivore damage and mortality in the period from March to July, but the latter was recorded as an additional variable. All statistical analyses were conducted in R (R Core Development Team, 2011) with the *nlme* package (Pinheiro and Bates, 2000) for Gaussian data and with the *lme4* package for binary data (Crawley, 2007, p. 605). Blocks and plots were treated as random effects, with plot nested in string. Fire and fertilization treatments were treated as random effects.

3. Results and discussion

There were 223 seedlings at the start of the experiment in November 2010. Of these, 152 (68%) survived the first three months, i.e., established successfully after transplantation. Survival was not related to fire history or fertilization treatments. Only 15% of the seedlings had signs of insect herbivore damage, so the "moderate" and "severe" categories were pooled and herbivory was analyzed as a binary response variable. There was no relationship between herbivore damage and any of the treatments. For the seedling pool surviving transplantation, there was no effect of fire history on any of the variables tested (shoot and root mass, root:shoot mass ratio, or SLA; Table 1, Fig. 1). Root mass and the root:shoot ratio responded weakly to P addition and the N:P interaction, with the P treatments leading to lower absolute and relative allocation to root tissue (Table 1, Fig. 1). There was no effect of fertilization on shoot biomass or SLA (Table 1).

The results clearly show that, at least over the duration of the experiment, there was no accumulated effect of fire history on the ability of Kruger soils to influence woody plant survivorship and growth. This result is consistent with past work showing that levels of plant-available N and P are not related to fire history in this system, except indirectly through the effects of fire on canopy cover (Coetsee et al., 2010, 2008; Holdo et al., 2012). Since all soil samples were collected in open sites away from tree canopies, there is no reason to believe that differences in tree cover may have played a role in the analysis. A potential concern with bioassay studies involving trees is that much of the initial growth may be driven by seed energy and nutrient reserves rather than by uptake from the soil. *Combretum hereroense* seeds collected for the experiment and set aside for seed mass calculations were found to have a mean mass of 0.1 g (N = 6), or about a fifth of the combined root and

Table 1
Results on mixed-model ANOVA of soil origin (defined by fire treatment at each of three strings or experimental burn blocks) and fertilization effects on shoot and root biomass, root:shoot biomass ratios, and specific leaf area (SLA) of *Combretum hereroense* seedlings grown in pots in the Skukuza Nursery, Kruger National Park. Values in bold are significant at the $\alpha = 0.05$ level.

Factor	d.f.		F and P-values							
	Model	Error	Shoot		Root		Root:shoot		SLA	
			F	P	F	P	F	P	F	P
Soil origin	2	4	1.33	0.36	0.07	0.93	2.92	0.17	0.24	0.80
N	1	118	0.11	0.74	0.08	0.78	0.07	0.80	2.03	0.16
P	1	118	0.01	0.91	2.26	0.14	4.13	**0.044**	0.09	0.76
N × P	1	118	0.68	0.41	4.10	**0.045**	3.63	0.059	0.39	0.53
Soil × N	2	118	0.31	0.73	0.41	0.66	0.42	0.66	0.44	0.64
Soil × P	2	118	0.68	0.51	0.56	0.57	0.22	0.80	1.52	0.22
Soil × N × P	2	118	0.03	0.97	0.66	0.52	0.90	0.41	0.23	0.79

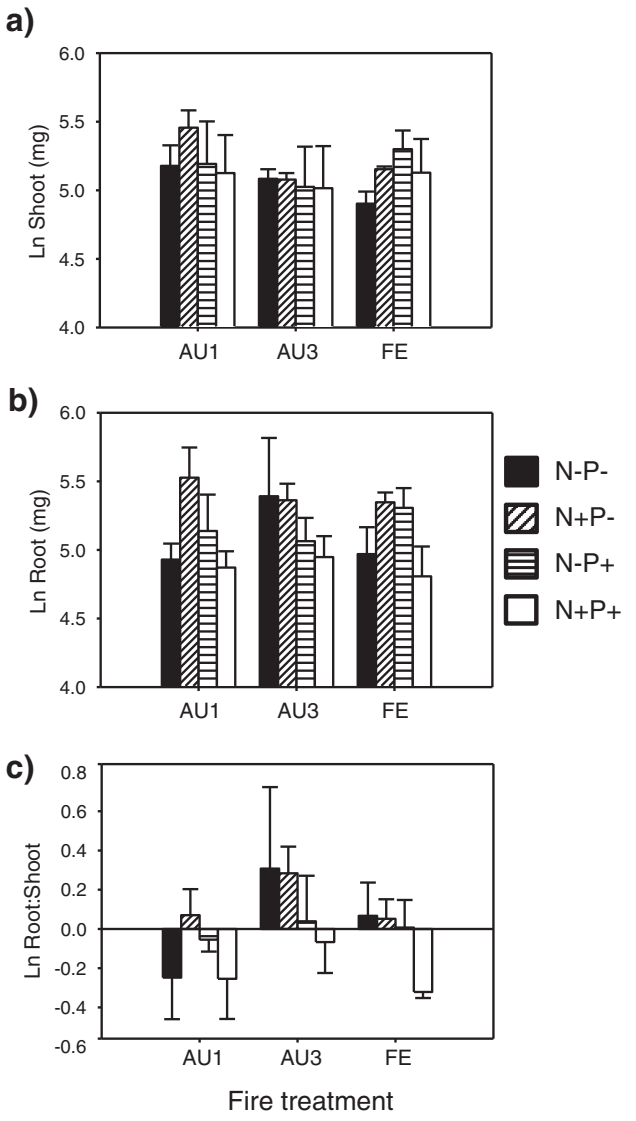

Fig. 1. Soil origin (as defined by fire treatment: FE = unburned; AU1 = annual August burns; AU3 = triennial August burns) and N- and P-fertilization effects on *Combretum hereroense* seedlings grown in pots in the Skukuza Nursery, Kruger National Park: a) Shoot mass, b) root mass, and c) root:shoot mass ratio seven months after transplantation.

shoot biomass of the harvested seedling. Though not insignificant, seed effects could only have contributed less to variance in growth than soil resources.

A previous fertilization experiment conducted on savannah grasses in KNP found synergistic effects of N and P addition on aboveground growth, suggesting that plants in this system respond non-additively to N and P (Craine et al., 2008). In contrast, other work conducted at multiple sites in nutrient-poor savannahs found no vegetation biomass response to fertilization by N or P, despite clear fertilization effects on plant-available P in the soil and foliar P (O'Halloran et al., 2010). Even when foliar N and P do respond to soil additions in savannah systems, these values are difficult to interpret and may not necessarily reflect the degree of N, P, or co-limitation (Craine et al., 2008). The present study suggests that plant-available N and P in soils may exert interactive effects on woody species, but these effects are manifested primarily below ground. The trends in Fig. 1c suggest that the addition of P leads to a reduction in below-ground investment, which would be consistent with P limitation. The design of the Kruger experimental burn trials by necessity (given that it is a landscape-level experiment) limits the degree of replication of the various treatments. There is little support for the notion that fire alters the degree to which soil resources limit tree growth in this system, but there is evidence for a role of soil nutrient availability in tree demography, which should be explored further independently of the role of fire. To date there is little evidence therefore that fire degrades or alters soil chemistry in a way that is likely to influence vegetation structure and dynamics in nutrient-poor savannah ecosystems, despite extensive aboveground effects on vegetation biomass (Bond et al., 2005; Staver et al., 2011). This has important consequences for our understanding of and ability to model long-term vegetation dynamics, because it means that the impact of fire in these ecosystems is short-term, and that it is unlikely to lead to lagged effects on system dynamics via slow-acting mechanisms, such as reduced future growth as a result of degraded soil fertility.

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